

## HYPERTENSION

## GW26-e1335

**siRNA Inhibits AT2 Receptor In decreasing NO Generation by Recombinant Human Angiotensin converting Enzyme 2 in Cardiac Microvascular Endothelial Cells**

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**OBJECTIVES** siRNA was used to silence AT2 receptor to explore the effect of Ang (1-9) -ACE2-AT2 pathway on NO formation after the impact of recombinant human Angiotensin Converting Enzyme 2 (rhACE2) on the cardiac microvascular endothelial cells (CMVEC).

**METHODS** Human cardiac microvascular endothelial cells (CMVEC) were cultured in vitro and grouped as follows: ① The control group: normal CMVEC; ② AngII intervention group: on the basis of the control group, AngII ( $1 \times 10^{-6}$  mol / L) was added and incubated 24h; ③ On the basis of AngII intervention, rhACE2 was added for incubation 5, 10, 15, 30, and 60 min respectively; ④ AT2 receptor inhibitor group: based on AngII intervention, AT2 receptor inhibitor (10 μmol / L) was added for incubation 30min, and then rhACE2 (100 μmol / L) was added 30min. ⑤ AT2 siRNA transfection group: siRNA was used to transfect CMVEC, and Western blot to detect protein expression of AT2 receptor and the transfection efficiency after its transfection, and the highest transfection efficiency group was elected and given AngII intervention for 30min, and then rhACE2 (100 μmol / L) was added for incubation 30min. Also a negative siRNA control group (negative control, NCsiRNA) was set up: after NCsiRNA transfection, it was treated as described above. Griess reagent measurement was applied to detect NO content in cell culture supernatant, RT-PCR to detect the expression of eNOS mRNA in HUVEC, Western blot to detect the expression of phospho-eNOS. NO fluorescent probe DAF-FM DA was loaded to detect intracellular NO formation and the activity of endothelial nitric oxide synthase (eNOS).

**RESULTS** The content of NO in AngII intervention group ( $3.495 \pm 0.362$  nmol / L) was significantly lower than that in the control group ( $11.513 \pm 0.392$ ) ( $P < 0.05$ ). After rhACE2 treatment, the NO contents and the phosphor-eNOS expression levels of cultured cell liquid in subgroups were significantly higher than those in AngII intervention group ( $P < 0.05$ ). However the protein expression levels of eNOS mRNA and non-phospho-eNOS showed no significant difference compared with AngII intervention group ( $P > 0.05$ ). And after CMVEC was intervened by AT2 pathway inhibitor (PD123319), the expression levels of phosphor-eNOS were significantly lower than those in rhACE2 30min treated group ( $P < 0.05$ ). After the successful transfection of siRNA into CMVEC, Western blot test results showed that 48 h after transfection, the protein expression of AT2 receptor decreased ( $P < 0.05$ ). Compared with non-transfected control group and negative control group, eNOS activity and NO levels of the AT2 siRNA transfected group were significantly reduced.

**CONCLUSIONS** Ang (1-9) -ACE2-AT2 signaling pathway is important in rhACE2's promotion of the activity of human cardiac microvascular endothelial cell eNOS and the NO formation.

## GW26-e4448

**A method to estimate 24-hour sodium excretion from spot urine samples and the application for target-organ damage**

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**OBJECTIVES** 24 h urine sodium excretion is considered the most reliable method to evaluate the salt intakes. However, this method is cumbersome. So we want to exploit formulas to estimate 24-h urinary sodium using spot urinary samples in Chinese hypertensive population and explore the application value of this method in salt intake assessment and target organ damage.

**METHODS** 1. We enrolled 510 cases of hospitalized patients with hypertension, 2/3 of them were arranged randomly to formula group to develop a new formula and the remains were used to test the performance of the formula. All participants were instructed to collect a 24-h urine sample, a second morning voiding urine sample (SMU), and a post-meridiem urine sample in the late afternoon or early evening, prior to the evening meal (PMU). All samples were sent to measure sodium and creatinine concentration.

2. We compared the differences of office blood pressure, 24 hour ambulatory blood pressure and left ventricular hypertrophy, vascular stiffness and urine protein among groups of different sodium intake.

**RESULTS** 24 hour sodium excretion formulas was obtained using SMU and PMU respectively, which have good consistency. The difference between the estimated and measured values in sodium excretion is 12.66 mmol/day (SMU) and 9.41 mmol/day (PM), to be equal to 0.7 g (SMU) and 0.6 g (PM) salt intake. Comparing with Kawasaki and Tanaka method, the new formula shows the lower degree of deviation, and higher accuracy and precision. Blood pressure of high urinary sodium group is higher than that in low urinary sodium group ( $P < 0.05$ ). Left ventricular hypertrophy and urinary albumin / creatinine aggravated with the salt intake increase, this has eliminated the influence of other factors. All of morphologies of the relationship between ambulatory arterial stiffness index, pulse wave velocity and carotid intima-media thickness with quartiles of sodium intake resembled a J-shaped curve.

**CONCLUSIONS** In Chinese hypertensive population, the formulas to estimate 24 h urinary sodium using spot urinary samples spot urine are considered useful for estimating the mean level of population salt intake, and have a role in evaluating target organ damage.

## GW26-e0487

**Relationship between Blood Pressure Circadian Rhythm and Early Renal damage in the patients with Primary Hypertension**

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**OBJECTIVES** To investigate the relationship between blood pressure circadian rhythm and early renal injury for the patients with primary hypertension.

**METHODS** A total of 225 hypertensive patients were divided into two groups according to nocturnal blood pressure decline rate ( $<10\%$  into non-dippers and  $\geq 10\%$  into dippers). The nocturnal blood pressure decline rate, 24 h blood pressure (24h-PP) and blood pressure index (PPI) were determined according to the data from ambulatory blood pressure monitoring. The glomerular filtration rate (eGFR) was calculated by the MDRD and Cockcroft-Gault equations respectively. Fasting plasma glucose, BUN, Scr, Cys-C, TG, TC, LDL-C, HDL-C, UA and MAU were dynamically monitored and body mass index (BMI) was measured. The relationship between blood pressure circadian rhythm and early renal damage in the patients with primary hypertension was analyzed by using the univariate and multivariate regression methods. For all tests,  $P < 0.05$  was considered to be statistically significant.

**RESULTS** The non-dipper group ( $n=149$ ) has significantly lower eGFR level ( $80.6 \pm 21.8$  v.s.  $97.3 \pm 24.2$  mL/min by MDRD equation,  $P < 0.001$ ;  $70.4$  v.s.  $91.2$  mL/min by Cockcroft-Gault equation,  $P < 0.001$ ), but significantly higher MAU ( $15.6$  v.s.  $11.8$  mg/L,  $P = 0.012$ ) and Cys-C levels ( $1.0$  v.s.  $0.9$  mg/L,  $P = 0.006$ ) than the dippers ( $n=76$ ). Moreover, comparing to the dippers, the non-dippers with higher 24h-PP ( $56$  v.s.  $50$  mm Hg,  $P = 0.008$ ) and PPI ( $0.42 \pm 0.07$  v.s.  $0.39 \pm 0.06$ ,  $P < 0.001$ ) were inclined to arteriosclerosis. The multivariate correlation and logistic regression analyses demonstrated that the N-SBP was correlated to MAU; BUN, Cys-C and PPI were correlated to eGFR based on the calculation with MDRD equation; and the Cys-C, D-DBP, 24-DBP, UA and BUN were correlated to eGFR based on the calculation with Cockcroft-Gault equation.

**CONCLUSIONS** The behavior of the early renal injury was significantly different between the non-dipper and dipper groups, which indicates the abnormal circadian rhythm of blood pressure could increase the renal target organ damage.

## GW26-e1287

**Aortic stiffness is associated with the central retinal arteriolar equivalent and retinal vascular fractal dimension in a population along the southeastern coast of China**

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**OBJECTIVES** The objective of this study was to evaluate the association of the central retinal arteriolar equivalent (CRAE) and the retinal